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## CORRELATION BETWEEN SIZE OF THE FRUIT AND THE RESISTANCE OF THE TOMATO SKIN TO PUNCTURE AND ITS RELATION TO INFECTION WITH MACROSPORIUM TOMATO COOKE

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That artificial infection with *Macrosporium tomato* Cooke<sup>1</sup> from tomato on uninjured tomato fruit can be obtained, provided fruit of a certain maturity as measured by size is used, has been established by one of the writers.<sup>2</sup> The question naturally arises as to the cause of this apparent immunity or resistance in the fruit after it reaches a certain maturity.

Previous investigations along these lines have been recently reviewed and summarized by Hawkins and Harvey<sup>3</sup> as follows: "It is apparent that there is good evidence that some parasitic plants make their way into their host plants by breaking through the tissues mechanically. There is no doubt that some fungi secrete enzymes which break down the cell walls of certain plants and are thus able to make their way through the tissues of their hosts."

The same workers from their infection studies with *Pythium* on potato conclude: "There is considerable evidence that the main factor in this penetration is the growth pressure of the fungus filament, and the resistance of the white McCormick potatoes to this disease is due to cell walls that are more resistant to mechanical puncture than are the cell walls of extremely susceptible varieties."

The results obtained by Blackman and Welsford<sup>4</sup> in their studies with *Botrytis cinerea* on *Vicia Faba* are of special interest. They state that "the piercing of the cuticle is due solely to the mechanical pressure exerted by the germ tube as a whole or by the special outgrowth from it."

In the work reported in this paper the evidence obtained shows that:

i. While a chemical difference is found in the analysis of young and old fruits, this is not the limiting factor in infection with *Macrosporium*. The

<sup>1</sup> The fungus causing typical "nail-head" spots on tomatoes has been shown in a paper which is being prepared for publication to be different from *Macrosporium solani* E. and M. For reasons given there this *Macrosporium* should be referred to as *Macrosporium tomato* Cooke.

<sup>2</sup> Rosenbaum, J. *Macrosporium solani* on tomato fruit (Abstr.). *Phytopathology* 9: 51. 1919.

<sup>3</sup> Hawkins, L. A. and Harvey, R. B. Physiological study of the parasitism of *Pythium debaryanum* Hesse on the potato tuber. *Journ. Agr. Res.* 18: 275-297. 1919.

<sup>4</sup> Blackman, V. H., and Welsford, E. J. Studies in the physiology of parasitism. II. Infection by *Botrytis cinerea*. *Annals of Botany* 30: 389-398. 1916.

fungus grows just as readily on the pulp and extracts of old fruits as on those obtained from young tomatoes. Moreover, positive infection has been obtained on fruits of all degrees of maturity when the skin is injured or removed previous to infection.

2. Surface sections of old and young fruits failed to reveal the presence of stomata or other natural openings in the skin.

3. As the tomato fruit develops, the surface of the fruit changes from a dull to a shiny appearance. The chemical nature of this change has not been determined, but sections cut from old and young tomatoes show that the cuticle increases in thickness with the age of the fruit. The development of the cuticular layer may be at least a partial explanation of the resistance of mature fruit to infection. Dewdrops are more readily retained on the dull surfaces than on the shiny and mature surfaces.

4. The maturity of the fruit as measured by size is correlated with a definite resistance of the tomato skin to puncture. The latter may also be one of the limiting factors in securing infection with *Macrosporium* on tomato fruit.

The methods employed in arriving at these conclusions and the detailed data obtained in this connection were as follows:

The tomatoes were grown in a commercial way in the fields of southern Florida. The work was limited to one variety, the "Livingston Globe." At first fruits of various sizes were selected at random. In order to get a more accurate knowledge of the fruit used, a large number of blossoms were tagged and pickings for stabbing and inoculations were made from these tagged blossoms at the end of each week. In this way it was possible to tell exactly the age of the fruit used, from blossoming time until the fruit began to show color in the field.

The *Macrosporium* cultures used in this work were isolated from tomato fruit. They were kept in pure culture and spores obtained according to the method described by Kunkel.<sup>5</sup> In a few cases spores were taken from fruit naturally infected in the field.

The resistance of the skin of the fruit to puncture was determined by the use of the Joly balance as modified by Hawkins and Harvey (*l. c.*). The construction of the Joly balance need not be given, but certain modifications of the balance and the methods followed in calculating the results obtained in the use of this apparatus in the present work will not be out of place here.

In using this apparatus for determining the resistance of the tomato skin, a fine glass needle 78 microns in diameter fixed to a glass rod with wax was suspended from the bottom of the pan. This needle was used throughout the experiments except in one instance as indicated. The needle and rod were well within the capacity of the spring of the balance. In operation the

<sup>5</sup> Kunkel, L. O. A method of obtaining abundant sporulation in cultures of *Macrosporium solani* E. & M. Brooklyn Botanic Garden Mem. 1: 306-312. 1918.

tomato was placed on the stand of the instrument; the needle was lowered until it just touched the surface of the tomato and watched closely until a quick drop showed that it had penetrated the skin of the fruit. The reading on the scale of the instrument was then taken. From this reading the pressure required to puncture the tomato skin could be calculated.

To illustrate the method of calculation, let us suppose the scale reading at which penetration took place on a fruit to be 31.00. The upward pull of the stretched spring representing this number as the reading on the scale was determined by counterbalancing with weights placed on the pan. This pull equalled 7.42 grams. The entire weight of the glass rod, with the needle, was 12.04 grams. The pressure necessary to puncture the skin was, therefore, the difference between the downward force and the upward force, or the difference between 12.04 and 7.42, or 4.62 grams. The pressure necessary, therefore, to penetrate the skin of a particular fruit with a needle which was 78 microns in diameter amounts to 4.62 grams.

Where tomatoes of different sizes were used, a total of five fruits of each size were punctured. Since the hardness over the entire fruit varies somewhat, it was thought advisable to make part of the punctures around the style end and an equal number around the stem end. Generally ten punctures were made on each fruit. The average of these readings gives fairly accurately the pressure necessary to puncture a particular fruit. As a general rule, it was found that the stem end was slightly harder than the style end. The fruits were picked and brought into the laboratory where they were divided into two lots, each lot containing fruits of the same maturity. One lot was used for determining the resistance of the skin to puncture while the other lot was washed, placed in disinfected moist chambers, and inoculated by spraying with a suspension of *Macrosporium* spores. In addition to the inoculations made on fruits brought into the laboratory, additional inoculations were made in the field on fruits growing on the vines. In this case the fruit was sprayed with a suspension of spores and covered with a glazed paper bag for a few days. No difference in the amount of infection was obtained whether the fruit was inoculated in the

TABLE I. *Showing the Relation Between Resistance of the Skin to Puncture and Macrosporium Infection on Different Sized Tomatoes\**

Size	Color	Circumference in Inches	Average Weight in Grams	Pressure in Grams Necessary to Puncture Fruit (Average of 50 Stabs)	Percentage of Positive Infection with Macrosporium
<i>A</i> ....	Red	.....	.....	5.19	0
<i>B</i> ....	Green	10 3/4-11 1/4	254.87	5.87	0
<i>C</i> ....	Green	7 3/4- 8 1/4	115.12	5.70	0
<i>D</i> ....	Green	6 1/2- 6 3/4	66.36	4.08	37 1/2
<i>E</i> ....	Green	5- 5 1/2	34.22	3.52	85 5/7
<i>F</i> ....	Green	4- 4 1/2	18.17	3.26	72 8/11
		3- 3 1/2	7.39	2.66	100

\* Temperature of tomatoes when punctured, 23° C.

Needle used, 78 microns in diameter.

laboratory or in the field. While a large number of punctures and inoculations were made throughout the season, the results are so uniform that it will suffice to present in tabular form a few representative series.

In Table 1 are shown the results of picking at random; at the same time, fruits of various sizes. These were divided into two equal lots, one of which was used for puncturing to determine the resistance of the skin, while the other was used for inoculations.

Examination of this table shows that the resistance of the skin to puncture increases with the size of the fruit, and likewise that the amount of infection varies from 100% in the case of the smallest fruit to 37½% in the case of fruit approximately 5–6 inches in circumference, with no infection above that size. From this series, the point at which the hardness of the skin begins to show any appreciable effect on infection is approximately that at which 4.08 grams of pressure is necessary to puncture the skin.

Table 2 shows the results of puncturing and inoculating fruits of known age for seven consecutive weeks. The fruits used in these series were all tagged when in blossom. The data show, as in the preceding table, that the older the tomato the more resistant is the skin, and that the amount of infection decreases when the resistance of the skin to puncture is approximately such that 5.08 grams of pressure is necessary to puncture the fruit with a 78-micron needle.

TABLE 2. *Showing the Relation Between Resistance of the Skin to Puncture and Macrosporium Infection on Tomatoes of Different Age*

Age in Days	Color	Weight in Grams, Average of 10 Fruits	Equatorial Diameter in Centimeters, Average of 10 Fruits	Temperature at which Stabbing was Done	Pressure in Grams Necessary to Puncture Fruit (Average of 100 Stabs)	Percentage of Positive Infection with <i>Macrosporium</i> tomato
7*....	Green	0.24	0.70	..	0.97	100
14....	Green	6.74	2.30	21	2.99	100
21....	Green	64.66	5.18	25	4.21	85
28....	Green	82.37	5.40	22	4.90	49
35....	Green	95.10	5.46	21	5.08	23 1/3
41....	Green	147.91	6.55	21	5.96	0
48....	Green	91.86	6.92	23	6.74	0
55....	Turning	....	....	25	5.56	0
55....	Red	162.82	6.31	25	5.10	0

\* Size of needle used for 7-day-old fruit, 46 microns in diameter. At all other ages punctured a 78-micron needle was used.

Tagging of blossoms to obtain fruits of a known maturity has shown that in the majority of cases age of fruit is a better indication of maturity than is size. As would naturally be expected, not all the tomatoes in a given lot of fruits attain the same size in a given length of time. Such fruits, however, have a resistance of the skin in proportion to their age rather than to their size, and react accordingly when inoculated. For this reason then, it seems, resistance of the skin to puncture is a better index of maturity than is the size of the fruit. The former is also preferable in

predicting whether a certain tomato can or cannot be infected with *Macrosporium*.

#### SUMMARY.

In the development of a tomato fruit, the cuticular layer increases in thickness with the age of the fruit. Measurements to determine the resistance of the skin of tomatoes have shown that there is a definite and direct correlation between age and the resistance of the skin to puncture.

Infection experiments with *Macrosporium tomato* on tomato fruit have shown that the amount of infection which it is possible to obtain decreases with the age of the fruit.

While the results do not prove absolutely that the inhibition of infection is a purely mechanical one, the resistance of the tomato skin to puncture may explain, at least partially, the ease with which infection without previous injury is obtained on the young fruit but not on the older fruit.

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